

## Appendix C

## Tobamovirus RNA dependent RNA polymerase

A three-dimensional structure of a RNA-dependent RNA polymerase, that encoded by poliovirus, has been reported by Hansen et al. The alignment used for tree construction is enhanced below by the amino acid sequence of the poliovirus RdRP aligned to the tobamoviral RdRp's. A 3D line is provided that identifies the helices (upper case Roman) and beta-structure elements (numerals) described in that report. The footnote line refers to notes at the bottom that relate to functional or structural comments.

1	
<u>TMV-vul</u>	--MQFYYDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCI LDMSKSVA
<u>TMV-Rak</u>	--MQFYYDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCI LDMSKSVA
<u>TMV-WANG</u>	--MQFYYDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCI LDMSKSVA
<u>TOMV-L</u>	--MQFYYDKCLPGNSTLLNNYD---AVTMKLTDISLNVKDCI LDMSKSVA
<u>TMV-KR</u>	--MQFYYDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCI LDMSKSVA
<u>PMMV</u>	--MQYYYDKCLPGNSTILNEYD---AVTMQIRENSLNVKDCVLDFSKSVP
<u>TMGMV</u>	PDLQFYDVCLPGNSTILNKYD---AVTMRLRDNLSNVKDCVLDFSKSIP
<u>ORSV</u>	--MQFYYDTLLPGNSTILNEYD---AVTMNLRENNLNVKDCTIDFSKSVS
<u>TVCV</u>	--MQFYYDALLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>CR-TMV</u>	--MQFYNDTLLPGNSTILNEYD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>RMV-SH</u>	--MQFYYDTLLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>CRMV</u>	--MQFYYDTLLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>TMV-CG</u>	--MQFYYDTLLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>TMV-OB</u>	QDLQFYDYDKCLPGNSTVLNEFD---AVTMNCSDISLNVKDCVLDFSKSVP
<u>CGMMV</u>	--MQE FYDRCRCLPGNSFVLNDFD---AVTMRLRDNEFNLQPCRRTLSNLDP
<u>CGMMV-W</u>	TDMQE FYDRCRCLPGNSFVLNDFD---AVTMRLRDNEFNLQPCRRTLSNLDP
<u>CFMMV</u>	--MQNFYDACRCLPGNSFVLNDYD---SVTMRLVDNEINLQPCRRTLSKADP
<u>YCGMMV</u>	TDMQS FYDACRCLPGNSFVLNDYD---SVTMRLADNEFNLQPCRRTLSKADP
<u>SHMV</u>	--LQYFYDSWLPGNSFVQNNHD---QWSIISSDINLHSEAVRLDMNKRH-
<u>POLIORDRP</u>	GEIQWMRPSKEVGYPIINAPS KTKLEPSAFHY-VFEGVKEPAVLTKDPR
3D	-----
Footnote	11 1
TMV VUL	AP---KDQIK-PLIPMVRTAAEEMPRQTGLLENLVAMI KRN FNAPELSGII
TMV-RAK	AP---KDQIK-PLIPMVRTAAEEMPRQTGLLENLVAMI KRN FNAPELSGII
TMV WANG	AP---KDQIK-PLIPMVRTAAEEMPRQTGLLENLVAMI KRN FNAPELSGII
TOMV L	AP---KDVKP-TLIPMVRTAAEEMPRQTGLLENLVAMI KRN FNSPELGVV
TMV-KR	AP---KDQIK-PLIPMVRTAAEEMPRQTGLLENLVAMI KRN FNAPELSGII
PMMV	LP---RESET-TLKPVIRTAEKP RKP GLLENLVAMI KRN FNSPELGVV
TMGMV SPA	MP---KEVKP-CLEPVLR TAAEPPRAAGLLENLVAMI KRN FNAPDLTGTI
ORSV	VP---RQEE-FFTPVIRTAEPR SAGLLENLVAMI KRN FNAPDLTGTI
TVCV	LP---KEQPI-FLKP KIRTAEEMPRTAGLLENLVAMI KRN MNA PDLTGTI
CR-TMV	LP---KEQPI-FLKP KIRTAEEMPRTAGLLENLVAMI KRN MNA PDLTGTI
RMV-SH	VP---KERPV-FMKPKLRTAAEEMPRTAGLLENLVAMI KRN MNA PDLTGTI
CRMV	VP---KERPV-FMKPKLRTAAEEMPRTAGLLENLVAMI KRN MNA PDLTGTI
TMV-CG	LP---RERPI-FMKPKLRTAAEEMPRTAGLLENLVAMI KRN MNA PDLTGTI
TMV OB	LP---RDNTKVPMP TVIRTAEPR S QGLLENLVAMI KRN FN SPELGT V
CGMMV	VPALIKNEAQNFLIPV LRTACERPRIPGLLENLVAMI KRN MNT PDLAGTV
CGMMV-W	VPALVKSEAQNFLIPV LRTACERPRIPGLLENLVAMI KRN MNT PDLAGTV
CFMMV	VTESLKMEKEFLIPLGKTATERPRIPGLLENLIAIVKRN FNT PDLAGSL
YCGMMV	VAESIKLERKNIDKLDLKTATERPRIPGFLLENLVAIVKRN FNT PDLAGVL
SHMV	---IPRTKGEFLRPLLNTAVEPPRIPGLLENLLALIKRN FNAPDLAGQL
POLIORDRP	L---K-TNFEEAIFS KYVGNKITEVDEHMKEAVDHYAGQLMSLDINTEQ
3D	-----AAAAAAA-----
Footnote	
TMV VUL	DIENTASL VVDKFFDSYLLKE-----KRKPKNVSLFSRESLN RWLEKQE
TMV-RAK	DIENTASL VVDKFFDSYLLKE-----KRKPKNVSLFSRESLN RWLEKQE
TMV WANG	DIENTASL VVDKFFDSYLLKE-----KRKPKNVSLFSRESLN RWLEKQE
TOMV L	DIENTASL VVDKFFDSYLLKE-----KRKPKNFSLFSRESLN RWI AKQE
TMV-KR	DIENTASL VVDKFFDSYLLKE-----KRKPKNVSLFSRESLN RWLEKQE

PMMV	DIEDTASLVVDKFFDAYLIKE-----KKKP-KNIPLLSRASLERWIEKQE
TMGMV SPA	DIESTASVVVDKFFDSYFIKK-----EKYTKNIAGVMTKDSMMRWLENRK
ORSV	DIEDTAELVNVNKFWDAYIIDE-LSGGNVTPM-----TSDAFHRWMAKQE
TCV	DIEDTASLVVEKFWDSDYVDKE-FSGTNEMTM-----TRESFSRWLSKQE
CR-TMV	DIEDTASLVVEKFWDSDYIDKE-FSGTNEMTM-----TRESFSRWLSKQE
RMV-SH	DIEDTASLVVEKFWDAYVVKE-FSGTDGMAM-----TRESFSRWLSKQE
CRMV	DIEDTASLVVEKFWDAYVVKE-FSGTDGMAM-----TRESFSRWLSKQE
TMV-CG	DIEDTASLVVEKFWDAYVVKE-FSGTDGMAM-----TRESFSRWLSKQE
TMV OB	DMENTASVVADEFFDSYFLKDKLSCSLGDGGKNIIDRQALIRWMEKQE
CGMMV	DITNMSISIVDNFFSSFVRDE-V---LLDHLDLDCVRASSIQSFSDWFSCQP
CGMMV-W	DITNMSISIVDNFFSSFVRDE-V---LLDHLDLDCVRASSIQSFSDWFSCQP
CFMMV	DISSISKGVVNDNFFSTFLRDE-Q---LADHLCKVRSLSLFSAWFDNQS
YCGMMV	DIDTISKSVVNDNFFTTFLRDE-Q---LSDHLVRVRSCSLESFSAWFHQNQ
SHMV	DYDFLSRKVCDGFFGKLLPPD-VEASELLRIPVDHMYSVQNFDDWLKQE
POLIORDRP	MCLEDAMYGTDGLEALDLSTS-----AGYPYVAMGKKKRDILN-----
3D	-----
Footnote	
TMV VUL	QVTIGQLADFDVFVDPAVDQYR-HMYKAQPKQKLDTSIQTEYPA-LQTIV
TMV-RAK	RVTIGQLADFDVFVDPAVDQYR-HMIKAQPKQKLDTSIQTEYPA-LQTIV
TMV WANG	QVTIGQLADFDVFVDPAVDQYR-HMIKAQPKQKLDTSIQTEYPA-LQTIV
TOMV L	QVTIGQLADFDVFVDPAVDQYR-HMIKAQPKQKLDLSIQTEYPA-LQTIV
TMV-KR	QVTIGQLADFDVFVDPAVDQYR-HMIKAQPKQKLDTSIQTEYPA-LQTIV
PMMV	KSTIGQLADFDIFDLPAVDQYR-HMIKQQPKQRLDLSIQTEYPA-LQTIV
TMGMV SPA	EVLLDDDLANYNFTDLPaidaQYK-HMIKAQPKQKLDLSIQNEYPA-LQTIV
ORSV	KSTIRQLADFDVFVDPaidaQYK-HMIKAQPKQKLDLSPQDEYAA-LQTIV
TCV	SSTVGQLADFNFVDPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
CR-TMV	SSTVGQLADFNFVDPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
RMV-SH	SSTVGQLADFNFVDPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
CRMV	SSTVGQLADFNFVDPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
TMV-CG	SSTVGQLADFNFVDPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
TMV OB	KSTIGQLADYDFVDPaidaQYR-HIIKSQPKQKLDLSIQSEYPS-LQTIV
CGMMV	TSAVGQLANFNFIIDLPafDTYM-HMIKRQPKSRLDTSIQSEYPA-LQTIV
CGMMV-W	TSAVGQLANFNFIIDLPafDTYM-HMIKRQPKSRLDTSIQSEYPA-LQTIV
CFMMV	TCALGQLSNFDFVDPPLPPDVYN-HMIKRQPKSKLDTSIQSEYPA-LQTIV
YCGMMV	TAAMGQLANFDFSDLPPVDMYT-HMIKRQPKSKLDTSIQSEYPA-LQTIV
SHMV	PGVGQLANWDHIGMPAADQYR-HMIKRTPAKLDSLQSEYPA-LQTIV
POLIORDRP	KQTRDTKEMQKLLDTYGINLPLVTYVKDELRSKTKV-EQGKSRLIEASSL
3D	-----
Footnote	
TMV VUL	YHSKKINAIFGPLFSELTRQLLDSDVDRFLFFTRKTPAQIEDFFGDLDS
TMV-RAK	YHSKKINAIFGPLFSELTRQLLDSDVDRFLFFTRKTPAQIEDFFGDLDS
TMV WANG	YHSKKINAIFGPLFSELTRQLLDSDVDRFLFFTRKTPAQIEDFFGDLDS
TOMV L	YHSKKINAIFGPLFSELTRQLLDSDIDSSRFLFFTRKTPAQIEDFFGDLDS
TMV-KR	YHSKKINAIFGPLFSELTRQLLDSDSSRFLFFTRKTPAQIEDFFGDLDS
PMMV	YHSKKINALFGPVFSELTRQLLETIDSSRFMFYTRKTPAQIEDFFGDLDS
TMGMV SPA	YHSKQINGILAG-FSELTRLLLEAFDSKKFLFFTRKTPEQIQQEFDLDS
ORSV	YHSKQINAIFGPLFAELTRQLLERIDSSKFLFYTRKTPEQIQQEFDLDS
TCV	YHSKKINAIFGPMFSELTRMLLERIDSSKFLFYTRKTPAQIEDFFGDLDS
CR-TMV	YHSKKINAIFGPMFSELTRMLLERIDSSKFLFYTRKTPAQIEDFFGDLDS
RMV-SH	YHSKKINAIFGPMFSELTRMLLERIDTSKFLFYTRKTPAQIEDFFGDLDS
CRMV	YHSKKINAIFGPMFSELTRMLLETIDTSKFLFYTRKTPAQIEDFFGDLDS
TMV-CG	YHSKKINAIFGPMFSELTRMLLERIDTSKFLFYTRKTPAQIEDFFGDLDS
TMV OB	YHSKKINALFGPIFSELTRQMLSAIDTSRFLFFTRKTPEQIQQEFDLDA
CGMMV	YHPKVVNAVFGPVFKYLTTFKFLSMDSSKFFFYTRKKPEDLQEFFSDLSS
CGMMV-W	YHPKVVNAVFGPVFKYLTTFKFLSMDSSKFFFYTRKKPEDLQEFFSDLSS
CFMMV	YHSKLVNAVFGPVFRYLTSFLSMDNSKFFFYTRKLRMICKFLFPHFPN
YCGMMV	YHSKLVNAVFGPVFRYLTSFLSMDNSKFFFYTRKTPEDLQSFFSTLSA
SHMV	YHSKHVNAVFGPIFSCLTERLLSVDPLRFKFFTRTPADLEFFFRDMVV
POLIORDRP	NDSVAMRMAFGNLYAAFHK-----NPGVITGSAVGCDPDLFWSKIPV
3D	-----BBBBB-CCCCCCCC-----DDDDDD



CGMMV-W	CFKASFCGDDSLIYLPKGLEYPDI-QATA--NLVWNFEAK-----LFRKK
CFMMV	SFKAAFCGDDSLIYMPPNLEYNDI-QSTA--NLVWNFEAK-----LYKKK
YCGMMV	SFKASFCGDDSLIYMPPGLEYPDI-QATA--NLVWNFEAK-----LFKKR
SHMV	VFKAGFCGDDSLIYLPRNLLYPDI-QSVS--NNMWNFEAK-----LFKKL
POLIORDRP	HLKMIAYGDDVIASYPHEVDASLLAQSGKDYGLTMPADKSAIFETVTWE
3D	222222--333333----IIIIIIII-4444--
Footnote	44 9
TMV VUL	YGYFCGRYVIHHDRGCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
TMV-RAK	YGYFCGRYIIHHDRGCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
TMV WANG	YGYFCGRYVIHHDRGCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
TOMV L	YGYFCGRYVIHHDRGCIVYYDPLKL-ISKLGAKHIKDWDHLEEFRRSLCD
TMV-KR	YGYFCGRYVIHHDRGCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
PMMV	YGYFCGRYIIHHDRGCIVYYDPLKL-ISKLGAKHINKREHLEEFRTSLCD
TMGMV SPA	YGYFCGRYIIHHDKGAIYYDPLKL-ISKLGAKHIKDYDHLEELRVSLCD
ORSV	YGYFCARYIIHHDRGAIVYYDPLKL-ISKLGCKHIKSLDHLEEFRMSLCD
TVCV	YGYFCGRYVIHHDRGAIVYYDPLKL-ISKLGCKHIRDVHLEELRESLCD
CR-TMV	YGYFCGRYVIHHDRGAIVYYDPLKL-ISKLGCKHIRDVHLEELRESLCD
RMV-SH	YGYFCGRYVIHHDRGAIVYYDPLKL-ISKLGCKHIRDEVHLEELRRSLCD
CRMV	YGYFCGRYVIHHDRGAIVYYDPLKL-ISKLGCKHIRDEVHLEELRRSLCD
TMV-CG	YGYFCGRYVIHHDRGAIVYYDPLKL-ISKLGCKHIRDEVHLEELRRSLCD
TMV OB	YGYFCGRYVIHHDRGAIVYDPLKI-ISKLGAKHITDKEHLEEFRISLAD
CGMMV	YGYFCGKYIIHHANGCIVYPDPLKL-ISKLGKNSLVGYEHVEEFRISLLD
CGMMV-W	YGYFCGKYIIHHANGCIVYPDPLKL-ISKLGKNSLVGYEHVEEFRISLLD
CFMMV	YGYFCGKYVIHHANGCIVYPDPLKL-ISKLGKNSLESYDHLEEFRISLMD
YCGMMV	YGYFCGKYVIHSNGCIVYPDPLKL-ISKLGKNSLESYDHLEEFRISLMD
SHMV	HGYFCGRYILRNNGRYLRLLPDPLKI-ITKLGCKAIKDWDHLEEFRISMFD
POLIORDRP	NVTFLKRFFRADEKYPFLIHPVMPMKEIRESIRWTKDPRNTQDHVRSLCD
3D	-555--66666-----77777-JJJJJJJJ---KKKKKKKKKKKKKKKK
Footnote	77 88 8
TMV VUL	VAVSLNN-CAYYTQLDDAVWEVHTAPPGSFVYKSLVKYLSDKVLFRSLF
TMV-RAK	VAVSLNN-CAYYTQLDDAVWEVHTAPPGSFVYKSLVKYLSDKVLFRSLF
TMV WANG	VAVSLNN-CAYYTQLDDAVWEVHTAPPGSFVYKSLVKYLSDKVLFRSLF
TOMV L	VAESLNN-CAYYTQLDDAVGEVHTAPPGSFVYKSLVKYLSDKVLFRSLF
TMV-KR	VAVSLNN-CAYYTQLDDAVWEVHTAPPGSFVYKSLVKYLSDKVLFRSLF
PMMV	VAGSLNN-CAYYTHLNDAVGEVIKTAAPLGSFVYRALVKYLCDKRLFQTLF
TMGMV SPA	VACSLGNWCLGFQPQLNAAIKEVHTAIDGSFAFNCVNKFLCDKFLFRTLF
ORSV	VSSSLNN-CALFGQLNDAIAEVHKTAVNGSFAFCSIVKYLSD?????????
TVCV	VASNLNN-CAYFSQLDEAVAEVHKTAVGGSFAFCSIICKYLSDKRLFRDLF
CR-TMV	VASNLNN-CAYFSQLDEAVAEVHKTAVGGSFAFCSIICKYLSDKRLFRDLF
RMV-SH	VTSNLNN-CAYFSQLDEAVAEVHKTAVGGAFVYCSIICKYLSDKRLFKDLF
CRMV	VTSNLNN-CAYFSQLDEAVAEVHKTAVGGAFVYCSIICKYLSDKRLFKDLF
TMV-CG	VTSNLNN-CAYFSQLDEAVAEVHKTAVGGAFVYCSIICKYLSDKRLFKDLF
TMV OB	VSKSLNN-CAYYAQLDEAVREVHKTAPPGSFVYKCIKFLSNRVLFESLF
CGMMV	VAHSLFN-GAYFHLLDAIHELPNAGGCSFVINCLCKYLSDKRLFRSLY
CGMMV-W	VAHSLFN-GAYFHLLDAIHELPNAGGCSFVINCLCKYLSDKRLFRSLY
CFMMV	VAKPLFN-AAYFHLLDAIHEYFPSVGGSTFAISLCKYLSNQLFGSLF
YCGMMV	VAKPLFN-AAYFHLLDAIHEYFPSVGGSSFAINSLCKYLSDKWLFRSLF
SHMV	MACEYKN-CFGFDVLESAVKESFPKAEGCNVAFCAIYKFLSNKYLFRTLF
POLIORDRP	LAWNNGE-EEYNKFL-----AKIRSPIGRALLPEYSTLYRRWL
3D	KKKK--L-LLLLLL-----LLLL--MMMM-----NNNNNNNNNN
Footnote	8 0
TMV VUL	ID
TMV-RAK	IN
TMV WANG	ID
TOMV L	LD
TMV-KR	ID
PMMV	LE
TMGMV SPA	LN
ORSV	??
TVCV	FV

CR-TMV	FV
RMV-SH	FV
CRMV	FV
TMV-CG	FV
TMV OB	F?
CGMMV	ID
CGMMV-W	ID
CFMMV	IK
YCGMMV	AK
SHMV	SD
POLIORDRP	DS
3D	NN

Footnote

- Footnotes:
- 1. N-terminal residues interacting with the thumb.
- 4. Three aspartates involved in metal ion binding at the active site.
- 5. Determinant of preference for NTP over dNTP. Could interact with 2' OH of substrate NTP.
- 6. Caps a helical turn
- 7. beta-turn.
- 8. Interaction across interface I
- 9. Interaction across interface I with Arg 456
- 0. See 9.
- a. Extra residues are present between helix H and beta-2 for poliovirus and between helix D and helix C for tobamoviruses. Their sequence similarity (YKGID for polio vs. LDSID for ToMV) suggests that they may serve similar functions in providing part of Interface I.

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[Return to Home Page](#)

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